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Research Notes : United States : Variation in pollen receptivity in artificial crosses of ms1-Urbana line

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1) A new mutation at the *ms1* locus.

Five different populations have been recognized as sources of *ms1* alleles. Genetic studies of male-sterile, female-fertile mutations conducted by Palmer et al. (1978) showed that *ms1*-North Carolina (T260), *ms1*-Urbana (T266), *ms1*-Tonica (T267), and *ms1*-Ames (T268) are independent mutations at the *ms1* locus. Yee and Jian (1983) reported another mutation at the *ms1* locus, designated Shennong Male-Sterile Soybean L-78-387.

The objective of our study is to determine if a spontaneous mutation that occurred within progeny developed from AP6(S1)C1 population is associated with the *ms1* locus.

Materials and methods: One hundred S_1 seeds of AP6(S1)C1 were planted in the spring of 1979 in Ron Secrist's plant nursery, and 55 single plants were harvested that fall. Among $S_{4:5}$ hill plots, one hill plot segregated for sterility. Figure 1 shows the origin of this hill. Twelve $S_{5:6}$ progenies (8 of fertile, 4 of sterile plants) were grown in Ames in 1985. Classification for male sterility and fertility involved the stainability of pollen grains in I_2KI and pod set at maturity. Testcrossing was conducted by using homozygous recessive *ms1*-Urbana plants as female parents and heterozygotes of the new mutants as male parents. Sixty-nine F_1 seeds were obtained for the allelism test. Thirty-one F_1 seeds were grown during fall/winter 1985 at the UPR/ISU Soybean Breeding Nursery in Isabela, Puerto Rico. They were classified for male-sterility/fertility on the basis of pollen staining.

Results: The new mutations arose in a population characterized by very complicated nuclear genetic background. AP6(S1)C1 population was derived by intermating and recurrent selection procedure from 40 strains of Group 0 to Group IV maturity (Fehr and Ortiz, 1975). It is worth mentioning that in this same population a partially male-sterile mutant *msp msp* was found in 1974/75. Expression of *msp msp* genes influenced different flower size and morphology, anther and pollen appearance, and phenotype at maturity (Stelly and Palmer, 1980).

Unknown sterile mutants showed similarities to the pattern of abnormalities caused by the *ms1* alleles. They exhibited prolonged vegetative growth

and produced large coenocytic pollen grains. Sterile plants had approximately 5.1 pods, with 5.9 seeds. Sterile *msl*-Urbana plants had 7.1 pods, with 14.9 seeds.

Among eight single-plant progenies observed in 1985, six progenies segregated for sterility, two did not. Within segregating progenies, 392 plants were fertile, 140 plants were sterile; that fit a ratio of 3:1, chi-square = 0.4912, $P = 0.10-0.50$ (Table 1).

$S_{5:6}$ progenies of four sterile plants gave the ratio of 23 fertile to 32 sterile plants. These results pointed out that this spontaneous new mutation is inherited monogenically. Testcrosses between *msl*-Urbana and *Msl msl* unknown mutants confirmed our cytological observations of sterile plants. Sixteen F_1 plants had normal pollen, 15 F_1 plants were characterized by large, coenocytic pollen grains. This population gave a good fit to the expected 1:1 ratio, chi-square = 0.032, $P = 0.50-0.90$ (Table 1). Seeds of these 15 F_1 fertile plants will be planted in 1986 for further observations.

The results indicated that this mutation occurred independently and a single locus was conditioning male sterility. The gene responsible for male sterility is allelic to the *msl* locus.

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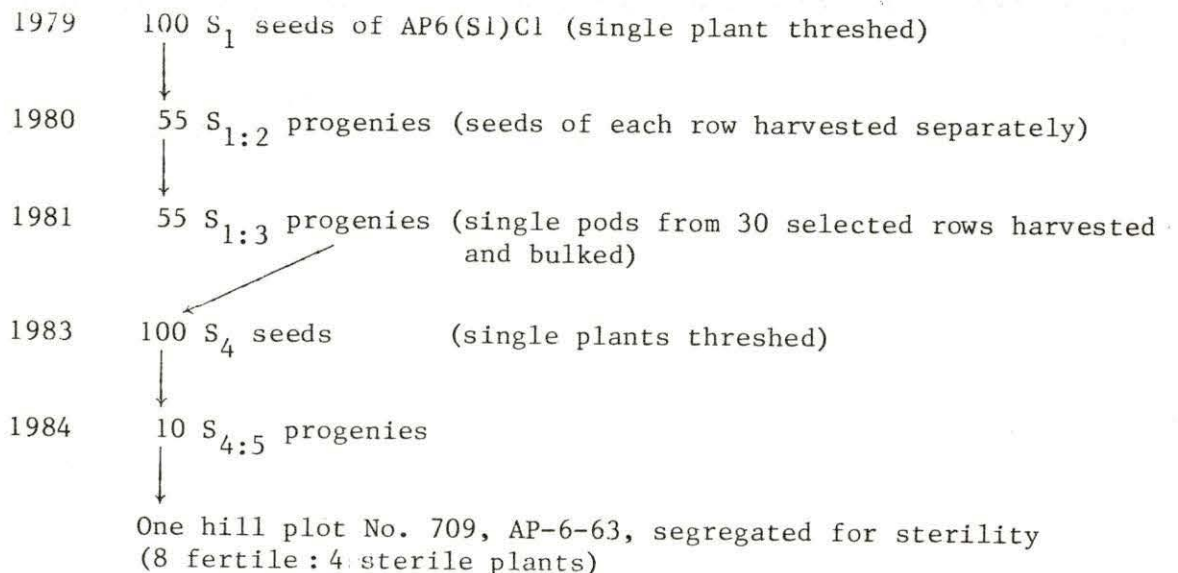


Figure 1. Origin of unknown sterile mutant

Table 1. Segregation ratios for fertility/sterility in $S_{5:6}$ progenies and F_1 testcross population

Parentage	Number of plants			Expected ratio	χ^2	P
	Observed	Fertile	Sterile			
$S_{5:6}$ progenies of fertile plants	532	392	140	3:1	0.4912	0.10-0.50
$S_{5:6}$ progenies of sterile plants	55	23	32			
F_1 (<i>ms1 ms1</i> -Urbana) x <i>Ms1 ms1</i> unknown mutant	31	16	15	1:1	0.032	0.50-0.90

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2) Test for apomixis in *ms4* male-sterile soybean.

Soybean plants homozygous for the male-sterile mutation *ms4* are capable of seed production in the absence of insect pollinators (Graybosch and Palmer, 1984). Cytological investigations have demonstrated the genesis of pollen grains by male-sterile plants at a frequency of 3.3% (Graybosch and Palmer, 1985). Pollen formed is identical to that of male-fertile plants, and will germinate when placed in an *in vitro* germination medium. A test using the genetic marker *y11* was designed to determine whether seed production by male-sterile plants was a function of the activity of these pollen grains, or apomixis. The *ms4* and *ms1* mutations both influence the process of postmeiotic cytokinesis during microsporogenesis. The *ms1* mutation also has a pleiotropic effect on female reproduction. A high frequency of polyembryonic and polyploid progeny results from the occasional omission of postmeiotic cytokinesis during megasporogenesis (Kennell and Horner, 1985). Since *ms1* and *ms4* demonstrate similar effects on male reproduction, it seemed possible that *ms4* also might influence female reproduction. Omission of cytokinesis during megasporogenesis could result in the formation of diploid eggs, followed by apomictic seed development. However, the recovery of polyploid and/or polyembryonic seedlings from *ms4* male-sterile plants has not been reported.

Male-sterile plants (*ms4 ms4 Y11 Y11*) were crossed with male-fertile plants heterozygous for *y11*. *y11* is a chlorophyll-deficient mutant; individuals heterozygous for *y11* display a yellow-green phenotype. Homozygosity for the recessive *y11* is a lethal condition. In the F_1 , male-fertile plants heterozygous for *y11* were selected and increased. F_2 seed was sown in single-plant progeny rows in the field in 1984 at Ames, Iowa. The F_2 population was rogued so that only individuals of the genotype *ms4 ms4 Y11 y11* remained. Seed production by these male-sterile plants was low, so F_3 seed was bulked for analysis. The F_3 was planted in 1985 at St. Charles, Missouri. Plants were classified for male sterility/fertility and for *y11*.

Seed produced by the F_2 male-sterile plants could have been the result of either self-perpetuation (self-pollination or apomixis) or outcrossing. Since fertile sibs were rogued from the F_2 , no male-fertile heterozygotes were available as donors of gametes carrying *ms4*. Thus, male-fertile individuals in the F_3 were the result of outcrossing; male-sterile plants only could have arisen via self-perpetuation. The classification of F_3 male-sterile plants

for *y11* is given in Table 1. If apomixis had occurred, all male-sterile individuals of the F_3 would have been of the genotype *Y11 y11*. If male-sterile progeny of male-sterile plants were the result of self-pollination, the resulting F_3 would have been composed of a population consisting of *1Y11 Y11:2Y11 y11:1y11 y11*. Since the *y11 y11* genotype is a seedling lethal, these plants could not be classified for male sterility. However, as the only sources of *y11* in the F_2 population were male-sterile plants, *y11 y11* individuals in the F_3 should have been of the genotype *ms4 ms4*. If *y11 y11* individuals are removed from consideration, self-pollination would be indicated by a 1:2 ratio of *Y11 Y11:Y11 y11*. Two chi-square analyses are given. Both fit the ratios expected if male-sterile progeny were the result of self-pollination.

Thus, the occurrence of apomixis is not supported by the results of this experiment. The test does demonstrate that the pollen grains formed by *ms4* male-sterile plants are capable of fertilization. There is no other explanation for the recovery of homozygous (*Y11 Y11* and *y11 y11*) individuals from male-sterile plants heterozygous for *y11*.

Table 1. Classification of F_3 male-sterile plants for the genetic marker *y11*

Genotype	N	Chi-square (1:2:1)	P	Chi-square (1:2)	P
<i>Y11 Y11</i>	79	2.786	>0.25	1.47	>0.10
<i>Y11 y11</i>	136				
<i>y11 y11</i>	60				

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3) Linkage tests with a locus conditioning ineffective nodulation response to *Rhizobium fredii*.

A single Mendelian locus has been described (Devine, 1984) that conditions ineffective vs. effective nodulation response upon inoculation of soybean lines with *Rhizobium fredii* (Scholla and Elkan, 1984). A formal gene symbol has not been assigned to this locus; for convenience, we will refer to it here as Rj?. In a breeding program with a long-term goal of developing adapted cultivars with an effective response to improved *R. fredii* strains, it might be more efficient to select for the effective nodulation response indirectly (via a tightly linked genetic marker) than by screening lines for nodulation response directly. In this study, we tested 5 loci for linkage with the locus conditioning effective nodulation.

The cross 'Evans' (ineffective, Rj? Rj?) x 'Peking' (effective, rj? rj?) was made at Ames in the summer of 1983. F₁ plants were grown during the winter in Puerto Rico and F₂ populations were grown at Ames in the summer of 1984. F₂ plants were classified for pubescence color (conditioned by the T-t gene pair) at maturity, then single-plant threshed to develop F₂ families. F₃ individuals were grown in growth boxes and inoculated with *R. fredii* strain USDA 193, as described elsewhere (Du Teau et al., 1986), to determine F₂ genotypes for the nodulation response locus (Rj? vs. rj? rj?). Isozyme analyses were performed as described by Griffin and Palmer (nd). The enzymes and loci assayed were: amylase activity (*Spl*); aconitase activity (*Aco4*,) (Griffin and Palmer, nd); diaphorase activity (*Dial*); superoxide dismutase activity (*Sod1*). The data are presented in Table 1. Chi square analyses of the data indicate no significant deviations due to linkage. We conclude that the effective nodulation locus (Rj?) is independent of the loci *Aco4*, *Dial*, *Sod1*, *Spl*, and *T*.

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Table 1. Segregation of a nodulation response locus (Rj?) and 5 other loci in the cross Evans (Rj? *Aco4-c Dial sod1 Spl-a t*) x Peking (rj? *Aco4-b dial Sod1 Spl-b T*)

	Expected proportion								
Rj? x	3	6	3	1	2	1	Total	χ^2	df
<i>Aco4</i>	22	44	15	3	9	8	101	3.45	2
<i>Spl</i>	16	46	19	3	9	8	101	1.90	2
<i>dial</i>	27	33	19	3	12	5	99	3.42	2
	Expected proportion								
Rj? x	9	3	3	1			Total	χ^2	df
<i>Sod1</i>	61	20	13	7			101	0.69	1
<i>T</i>	61	20	19	1			101	2.43	1

Segregation of Rj?:

81 Rj? : 20 rj? rj?; (3:1) = 1.46

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4) Allelism tests of T218H and T225H.

Genetic type T218M was found in 'Illini' in 1952 at Urbana, IL. T218H was derived from T218M by crossing a yellow branch as male parent with Illini. The F_1 plants were green. Segregation in the F_2 gave 3 green:1 yellow lethal plants. F_2 green plants gave ratios of 1 nonsegregating:2 segregating progenies in the F_3 generation, confirming monogenic inheritance (Palmer, 1978, unpublished). The recessive allele is carried as the heterozygote in T218H.

Genetic type T225M was found in 'Lincoln' in Iowa before 1955. T225H was derived from T225M. In the growth chamber, a few weak yellow plants flowered and were used as male parents onto Lincoln as female parent. One hybrid plant was obtained. The F_1 plant was green and segregation in the F_2 gave 3 green:1 yellow lethal plants. Confirmation of inheritance was obtained in the F_3 generation. The recessive allele is carried as the heterozygote in T225H (Sheridan and Palmer, 1975).

The objective of this study was to test for allelism between T218H and T225H.

Results: Reciprocal crosses were made by using green plants of T218H and T225H. Self-pollinated seed of plants used as parents were planted in the sandbench to determine the genotype of the parents, i.e., AA or Aa .

Data of F_1 hybrid plants from crosses between two heterozygous parents are given in Table 1. Yellow plants were observed in the F_1 in a ratio of 3 green:1 yellow lethal plants.

The F_1 green plants were field grown in Ames and were threshed individually. About half of the seed from each F_1 plant were planted in the sandbench to determine segregation for green and yellow plants. The remaining seed were planted in the field the following summer and were classified for plant color. Data were combined (Table 1). Segregation of pubescence color in the F_2 confirmed the hybridity of the cross.

Among all F_2 families of the cross T225H x T218H, there were 14 segregating:5 nonsegregating families ($\chi^2_{2:1} = 0.43$; $P = 0.75-0.50$). In the reciprocal cross, there were 8 segregating:3 nonsegregating families ($\chi^2_{2:1} = 0.21$; $P = 0.75-0.50$).

Five plants from each field-grown F_2 entry were threshed individually and 50 seed per entry were planted in the sandbench and segregating families were classified for plant color (Table 3).

Table 1. Ratio of green:yellow F₁ plants from reciprocal crosses of known heterozygotes of T218H and T225H (field data)

<u>T218H x T225H</u>			
<u>Green</u>	<u>Yellow</u>	<u>Chi square (3:1)</u>	<u>P</u>
12	4	0.00	1.00
<u>T 225H x T218H</u>			
<u>Green</u>	<u>Yellow</u>	<u>Chi square (3:1)</u>	<u>P</u>
21	6	0.10	0.90-0.70

Table 2. Ratio of green:yellow F₂ plants from reciprocal crosses of known heterozygotes of T218H and T225H (greenhouse and field data combined)

<u>T218H x T225H</u>				
	<u>Green</u>	<u>Yellow</u>	<u>Chi-square (3:1)</u>	<u>P</u>
Totals	2059	636	17.70	
Pooled chi-square (1 df)			2.83	>0.10
Homogeneity chi-square (7 df)			14.87	>0.05
<u>T225H x T218H</u>				
	<u>Green</u>	<u>Yellow</u>	<u>Chi-square (3:1)</u>	<u>P</u>
Totals	3183	957	50.01	
Pooled chi-square (1 df)			7.84	>0.005
Homogeneity chi-square (13 df)			42.17	>0.005

Table 3. Ratio of green:yellow F₃ plants from reciprocal crosses of known heterozygotes of T218H x T225H (greenhouse data)

<u>T 218H x T225H</u>				
	<u>Green</u>	<u>Yellow</u>	<u>Chi-square (3:1)</u>	<u>P</u>
Totals	899	272	15.36	
Pooled chi square (1 df)			1.96	>0.20
Homogeneity chi-square (23 df)			13.40	>0.95
<u>T225H x T218H</u>				
	<u>Green</u>	<u>Yellow</u>	<u>Chi-square (3:1)</u>	<u>P</u>
Totals	1783	482	32.14	
Pooled chi-square (1 df)			16.72	>0.005
Homogeneity chi square (42 df)			15.42	>0.995

In the cross of T218H x T225H, the number of F_2 entries evaluated in the F_3 was 40. The number of segregating entries was 24:16 nonsegregating (χ^2 2:1 = 0.9; $P = 0.50-0.25$). In the reciprocal cross, the number of segregating entries was 43:26 nonsegregating (χ^2 2:1 = 0.73; $P = 0.5-0.25$).

Discussion: If T218H and T225H were allelic, one fourth of the F_1 plants would be yellow lethal. If these loci were nonallelic, all F_1 plants would be green. In Table 1, the data from reciprocal crosses show that one-fourth of the F_1 hybrid plants were lethal. This suggests that T218H and T225H are allelic.

The F_2 segregation indicated that two-thirds of the green F_1 plants were heterozygous and one-third homozygous dominant. Among segregating entries of the reciprocal crosses, a ratio of 3.24 green:1 yellow for T218H x T225H and 3.33 green:1 yellow for T225H x T218H was calculated. In both cross combinations, there was a deficiency of yellow plants.

The F_3 segregation also gave a 2:1 ratio for heterozygous:homozygous dominant genotypes. For 218H x T225H, we observed 3.31 green:1 yellow and for T225H x T218H, 3.70 green:1 yellow. Again, for both cross combinations, there was a deficiency of yellow plants.

In the reciprocal crosses in the F_2 and F_3 , we observed all green plants or segregation approximating 3 green:1 yellow. Furthermore, we never observed a 9 green:7 yellow, which would indicate two nonallelic loci.

The significant deviations from 3:1 within and among families of the reciprocal crosses has been noticed previously in crosses with T225H (Palmer, 1984).

We believe that these data confirm the allelism of these two mutants. The event(s) that produced the unstable allele in T218HM and T225M occurred in different cultivars and in different years. We conclude that they represent two independent events.

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5) An additional beta-amylase mobility variant conditioned by the *Sp1* locus.

The *Sp1* locus (Orf and Hymowitz, 1976; Gorman and Kiang, 1978) codes for beta-amylase (Hildebrand and Hymowitz, 1980). Two mobility variants and two null variants have been described (Orf and Hymowitz, 1976; Kiang, 1981). We have been conducting a survey of isoenzyme variability in the soybean germplasm collections. We assay beta-amylase activity on 10% acrylamide gels (Davis, 1964) to determine genotypes at the *Sp1* locus. A recent plant introduction from China (PI 464918) was found to have a beta-amylase with mobility intermediate to those of the *Sp1-a* and *Sp1-b* alleles.

This new variant was tested in crosses with lines carrying either the *Sp1-a* or *Sp1-b* alleles. The data are presented in Table 1. Both populations segregated in a 1:2:1 ratio as tested by chi-square, indicating that the new variant is conditioned by an allele of the *Sp1* locus. We designate the allele *Sp1-c*. The relative mobility of the three alleles is: *Sp1-a*, $R_f=.27$; *Sp1-c*, $R_f=.30$; *Sp1-b*, $R_f=.33$.

Table 1. Allelism tests of an additional beta-amylase mobility variant at the *Sp1* locus

Cross	Generation	Classes			N	χ^2
		<u>a/a</u>	<u>a/c</u>	<u>c/c</u>		
'Evans' x PI 464918						
a/a x c/c						
(includes reciprocal)	F ₂	120	186	104	410	4.75
PI 257430 x PI 464918						
b/b x c/c	F ₂	25	62	29	116	0.83

PI 464918 came into the U.S. under the name 'Ji Ti 4' from Heilongjiang Province, The People's Republic of China. Three other PIs from the same collection, 464916 ('Ji Ti 2'), 464917 ('Ji Ti 3'), and 464919 ('Ji Ti 5') were assayed to determine their genotypes at the *Sp1* locus. None had the *Sp1-c* allele. Further, in our survey of approximately 1500 accessions in the soybean (*Glycine max*) and wild soybean (*G. soja*) germplasm collections, PI 464918 is the only accession determined to have the *Sp1-c* allele. The 'Ji Ti' series are all improved cultivars. The absence of the *Sp1-c* allele in any other accessions indicates that this is probably a relatively recent mutation at the *Sp1* locus.

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6) Enhancing seed set in *Glycine falcata*.

Research using wild relatives of the soybean, such as *Glycine falcata* Benth., a native of Australia, has been hampered by an extremely low seed set under greenhouse conditions in the absence of pollinating vectors. A quick and efficient method of enhancing seed set in this species was developed in working with *Neonotonia verdcourtii*, a species from Africa formerly included in the genus *Glycine* (Isely et al., 1980). Conventional soybean crossing techniques among flowers of one plant were tedious and time-consuming, and did not produce many seeds.

We found that a tool with a rough tip served better than a sharp forceps to move pollen from flower to flower. Such a tool seems always to be at hand in dried petioles from fallen leaves. A slim stiff petiole inserted between the two keel petals and moved across the stamens gathers pollen. Moving from flower to flower, quickly rubbing this make-shift tool across the stamens, results in production of an abundance of seeds. No care is needed as to which flowers are treated; the procedure can go from branch to branch within the same plant or between plants. A ten-minute visit to the plants each day to treat the new flowers is sufficient to produce good seed set.

Reference

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H. E. Heer

7) Variation in pollen receptivity in artificial crosses of *msl*-Urbana line.

General characters associated with the soybean male-sterile *msl* gene have been described in a previous study (Chen et al., 1985). Among the four spontaneous and independent *msl* source populations (Palmer et al., 1978), the Urbana male-sterile (*msl*-Urbana) line was reported to have 1) higher female fertility (Boerma and Cooper, 1978), 2) lower percentage of ovule abortion (Kennell and Horner, 1985), and 3) higher percentage of pollen-tube germination in male-sterile anthers (Chen et al., 1986). However, no report has mentioned whether gynoecia of male-sterile plants would have the same receptivity as those of male-fertile plants, when fertile pollen was applied to the stigma of male-sterile plants.

In a study of the germination and growth of male-fertile pollen on gynoecia of male-fertile homozygous *Msl Msl*, male-fertile heterozygous *Msl msl*, and male-sterile homozygous *msl msl* plants, we used the *msl*-Urbana line. Gynoecia of male-sterile plants allowed the germination and growth of fertile pollen grains as well as those of male-fertile plants. Pollinations from fertile heterozygous plants onto fertile heterozygous gynoecia, however, resulted in a lower percentage of pollen-tube growth than did the other combinations (Chen, 1985). Further investigations during different growing seasons in greenhouse plantings were conducted. In this study, we report variation in pollen-tube germination and growth in gynoecia of artificial crosses with fertile pollen among the *msl*-Urbana progeny.

Materials and methods: Four greenhouse plantings, 1984 summer (1984 S), 1985 winter (1985 W), 1985 early summer (1985 SI), and 1985 middle summer (1985 SII) were grown.

A sibling male-fertile line not segregating for male sterility was used as the homozygous male-fertile *Msl Msl* (FH) plant source. Seeds obtained from male-sterile plants produced either male-fertile *Msl msl* (Fh) or male-sterile *msl msl* (SH) plants. Plants were classified as male fertile or male sterile at the beginning of flowering on the basis of pollen grain stainability with I₂KI. Different combinations of cross-pollinations were made among these three genotypes. Artificial crosses were made in all combinations during the same time period, but varied in number of crosses for each combination. Gynoecia were collected from each artificial crossing combination 24 hours

after pollination and fixed in FAA for 24 hours. Fixation, clearing procedure, and staining were the same as described by Chen et al. (1986). Observations were made by using light and fluorescence microscopy.

Results and discussion: Observations of pollen-tube growth could be grouped into three categories: normal pollen-tube growth, pollen-tube growth retarded or pollen tubes degenerated in stigma or style areas, or no observable pollen-tube growth. Percentage of gynoecia with normal pollen-tube growth became our primary interest because of its possible indication of fertilization. Types of crosses, as well as percentage of gynoecia with normal pollen-tube growth among different seasons of greenhouse plantings, are shown in Table 1. Variation in percentage of gynoecia with normal pollen-tube growth from season to season was noted. Although consistency in variation from cross to cross was not observed, Figures A-F indicate that variation from environment to environment in crosses of FH x FH vs. FH x Fh, and SH x FH vs. SH x Fh follow the same pattern (Figures A, B, E, and F). Nevertheless, crosses of Fh x FH vs. Fh x Fh (Figures C and D) did not show the same pattern as those of the others. Whether these results suggest a heterozygote effect on the female gynoecia for pollen receptivity is not known.

The results of the 1984 summer study indicated that male-fertile pollen grains germinated and grew readily in the style of the male-sterile plants. However, in the Fh x Fh cross, normal pollen-tube growth in the gynoecia was observed in only 54% of the artificial crosses. This compares with 88%, 91%, and 84% for the FH x FH, SH x FH, and SH x Fh crosses, respectively. A chi-square test among the FH x FH, Fh x Fh, SH x FH, and SH x Fh crosses indicated distribution of percentage of gynoecia with normal pollen-tube growth, pollen-tube degeneration, and no observable pollen-tube growth is significantly different at the 5% level. Most of the variation is contributed from the Fh x Fh crosses.

The artificial crossing study was repeated in winter 1985, early summer 1985, and middle summer 1985 to determine if there was any heterozygote effect on cross compatibility. Definite conclusions cannot be made from these studies. Nevertheless, from the overall data, equivalence or superiority of pollen from the FH plants, compared with that of the Fh plants, was noticed when different pollen sources were pollinated onto the gynoecia of the same genotype. This was noted from all crosses among environments, except the Fh x FH vs. Fh x Fh crosses in 1985 W (Table 1, underlined). The average

percentage of gynoecia with normal pollen-tube growth from all growing seasons varied from 73.0% to 57.2% with the sequential order $FH \times FH > Fh \times FH > SH \times FH > FH \times Fh > Fh \times Fh > SH \times Fh$ (Table 1). However, differences in percentage of gynoecia with normal pollen-tube growth are statistically non-significant in most cases. The success rate in hand pollination is known to vary from person to person and from environment to environment (Fehr, 1978). In our study, different cross combinations were made the same day with a varying number of crosses by the same person. Thus, if the differences in percentage of pollen-tube growth between the paired crosses (the same female genotype crossed with FH or Fh pollen) varied randomly, both positive and negative differences ($\times FH$ vs. $\times Fh$) should be expected with equal probability among these paired comparisons. As shown in Table 1, however, only one pair of crosses showed a negative difference (Table 1, underlined) whereas nine of the others showed no or positive differences. Differences in stigma and style receptivity have not been reported in soybean. In maize and other species, gamete competition between self-pollination and cross-pollination has been addressed (Johnson and Mulcahy, 1978; Yamada and Murakami, 1983; Ottaviano et al., 1983; Currah, 1983; Sarr et al., 1983). In our study, variation from cross to cross was noted. From the combined data, it seemed that $FH \times FH$ had the highest average percentage (73.0%) of pollen-tube growth, while that of the $SH \times Fh$ had the lowest (57.2%). An average of 9% difference in percentage of pollen-tube growth between pollen from FH plants and Fh plants was observed in gynoecia of the same female genotype. Although variation observed in our study was not supported by statistical analysis, there is a general trend. Furthermore, the environmental effect on expression of pollen-tube growth in gynoecia was significant. Thus, variation in the percent of pollen-tube growth in gynoecia may be influenced by environmental factors, genotypes, or interaction of both. Sorrells and Bingham (1979) indicated that the *ms1* allele had an effect on microspore cytokinesis in some fertile *Ms1 ms1* F_1 plants. Whether the inconsistent results obtained in this study are due to the unstable expression of the *ms1* allele in the heterozygous condition is not known. Further research might clarify whether this variation is controlled by factors such as the compatibility for pollen-tube growth in gynoecia or the instability of the *ms1* allele.

Table 1. Percent of gynoecia with pollen-tube growth among crosses of *ms1*-Urbana line grown in different environments

Environment ^b	Parental combinations ^a (♀ x ♂)						Average of crosses
	FH x FH	FH x Fh	Fh x FH	Fh x Fh	SH x FH	SH x Fh	
1984 S	87.5 (24) ^c	--	--	54.2 (24)	90.6 (32)	83.8 (37)	79.0 ± 8.4 (117)
1985 W	66.7 (21)	66.7 (21)	53.9 (39)	75.0 (32)	64.0 (25)	46.4 (28)	62.1 ± 4.2 (166)
1985 SI	52.0 (25)	50.0 (22)	68.8 (16)	38.8 (49)	41.4 (29)	27.3 (22)	46.4 ± 5.8 (163)
1985 SII	85.7 (63)	75.0 (60)	82.5 (57)	74.6 (55)	75.0 (44)	71.4 (56)	77.4 ± 2.2 (335)
Average of environments	73.0±8.4 (133)	63.9±7.4 (103)	68.4±8.3 (112)	60.6±8.8 (160)	67.8±10.4 (130)	57.2±12.8 (143)	65.1 ± 3.7 (781)

^aFH = Male-fertile homozygote, *Ms1 Ms1*;
 Fh = Male-fertile heterozygote, *Ms1 ms1*;
 SH = Male-sterile homozygote, *ms1 ms1*.

^bS = summer; W = winter; SI = early summer; SII = middle summer.

^c() indicates total number of crosses.

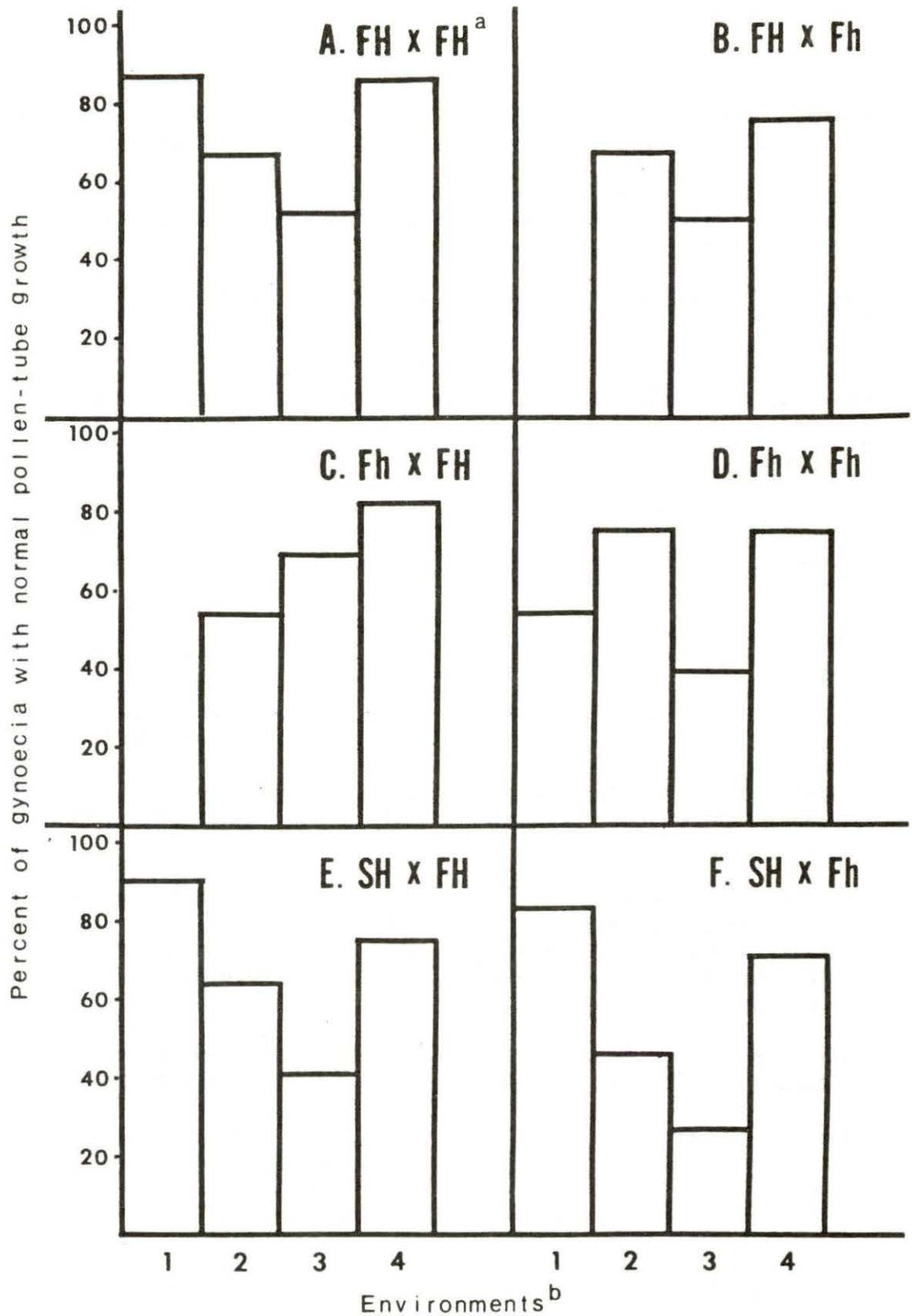


Fig. A-F: Percent of normal pollen-tube growth in gynoechia of artificial crosses of *ms₁*-Urbana progeny among different environments of greenhouse plantings.

a: FH: Male-fertile homozygote; Fh: Male-fertile heterozygote; SH: Male-sterile homozygote

b: Number in each column indicates environments, 1: 1984 summer; 2: 1985 winter; 3: 1985 early summer; 4: 1985 middle summer

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